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Acutely Lethal Levels of Cadmium, Copper, and Zinc to Adult Male Coho Salmon and Steelhead

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ABSTRACT

Flow-through acute toxicity tests of cadmium, copper, and zinc were conducted with adult male coho salmon (*Oncorhynchus kisutch*) and adult male steelhead (*Salmo gairdneri*). The 96-h LC50 values for copper were 46 and 57 μg/liter, and for zinc were 905 and 1,755 μg/liter, for coho salmon and steelhead, respectively. Mortality induced by cadmium was slow in onset, but 50% mortality occurred after more than a week at 3.7 μg/liter for coho salmon and 5.2 μg/liter for steelhead. Hardness and alkalinity of the water supply were higher during the toxicity tests with steelhead, complicating direct comparisons between the two species.

Anadromous salmonids return from the ocean as maturing adults in a state of progressive physical and physiological debilitation, manifested in fungal infections, skin lesions, organ atrophy, and cardiovascular changes. Debilitation is generally more severe in the Pacific salmon (*Oncorhynchus* spp.) which all die after spawning, than in the trout (*Salmo* spp.) which can return to the ocean (Robertson and Wexler 1960; Moore et al. 1976). The deteriorated state of adult salmonids could reduce their resistance to biological, physical, and chemical stresses. Juvenile salmonids represent one of the more pollution-sensitive forms of aquatic life for which toxicity data are available, and mature adult salmonids, because of their debilitation, may be even more sensitive to pollutants.

Few, if any, toxicity tests had been conducted on adult anadromous salmonids prior to this study because of their large size and the inherent problems of collecting and holding them. However, development of an adult salmon testing facility at the Western Fish Toxicology Station (WFTS) made such research practical, and led to this series of toxicity tests to determine the acute lethality of cadmium, copper, and zinc to adult salmonids.

METHODS

Coho salmon (*Oncorhynchus kisutch*) and steelhead (*Salmo gairdneri*), collected from hatchery holding facilities at the termini of their spawning migrations on the Alsea River in western Oregon, were transported in a refrigerated fish transport truck 32–64 km to the WFTS facility. Only male fish were used because females were required for hatchery spawning. Individual fish were captured, transferred to and from the truck, and distributed among test tanks in a smooth fabric brail to minimize netting injury. Fish were distributed among five identical 5,250-liter holding tanks (Chapman 1978a, this issue) by stratified random assignment.

Depending on the number of fish available, from 50 to 165 adult male coho salmon or steelhead were distributed among the five tanks. After allowing the fish up to 3 days for tank acclimation and recovery from transport and handling, four tanks were dosed with adequate metal stock solution to achieve the nominal concentration, and the stock solution flows were started. The fifth tank served as a control. Stock solution and water flow rates and turnover rates were identical to those previously used for adult salmon (Chapman 1978a, this issue).

Water for the tests was from a shallow (12 m) well on the bank of the Willamette River. Fluctuations in ambient temperature, hardness, and alkalinity occurred during the series of tests, but these fluctuations were merely recorded, and no control was attempted. Dilution water and stock solution flow rates were measured daily, and water temperature in each tank was recorded continuously. Metal stock solutions were prepared from reagent grade chloride salts ($CdCl_2 \cdot 2.5H_2O$; $CuCl_2 \cdot 2H_2O$; and $ZnCl_2$) dissolved in well water and dispensed from

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TABLE 1.—*Summary of basic data for toxicity tests of cadmium, copper, and zinc with adult male coho salmon and steelhead. Water chemistry data include range (in parentheses) and mean ± standard deviation.*

Species	Metal	Time (h)	LC50 and 95% confidence limits[a] (μg/liter)	Dissolved oxygen (mg/liter)	pH	Total hardness $CaCO_3$ (mg/liter)	Total alkalinity $CaCO_3$ (mg/liter)	Temp. (C)	No. fish per tank	Accli-mation time (days)
Coho salmon	Zn	96	905 (636–1,211)	9.8 ± 0.2 (9.5–10.0)	7.40 ± 0.28 (7.2–7.6)	25 ± 2 (23–26)	20 ± 7 (15–25)	13.7 (10.3–16.5)	10	0
Coho salmon	Cu	96	46 (44–49)	9.9 ± 0.2 (9.4–10.7)	7.29 ± 0.09 (7.2–7.4)	20 ± 1 (19–21)	22 ± 1 (21–22)	9.4 (8.7–11.1)	33	1
Coho salmon	Cd	215	3.7 (2.6–5.0)	9.8 ± 0.4 (9.0–10.4)	7.3 ± 0.04 (7.3)	22 ± 1 (20–23)	22 ± 1 (22–23)	10.0 (8.9–10.8)	20	1
Steelhead	Zn	96	1,755[b] (1,499–16,731)	10.4 (10.0–11.0)	7.45 ± 0.04 (7.4–7.5)	83 ± 8 (64–89)	55 ± 4 (45–59)	10.3 (7.6–12.4)	11	3
Steelhead	Cu	96	57[c] (46–68)	11.4 (10.6–12.0)	7.57 ± 0.06 (7.5–7.6)	42 ± 12 (28–59)	34 ± 8 (24–45)	9.2 (7.0–11.0)	11	2
Steelhead	Cd	408	5.2[d] (2.8–8.5)	10.7 (9.7–11.3)	7.50 ± 0.10 (7.4–7.6)	54 ± 6 (28–90)	39 ± 10 (25–62)	9.6 (6.9–11.5)	12	3

[a] Moving average method.
[b] 735 μg/liter when adjusted to a hardness of 20 mg/liter (after Brown 1968).
[c] 35 μg/liter when adjusted to a hardness of 20 mg/liter.
[d] 1.3 μg/liter when adjusted to a hardness of 20 mg/liter.

100-liter plastic containers. Typical water quality data for the well water are listed elsewhere (Chapman 1978b, this issue; Samuelson 1976).

Fish were routinely checked every few hours during daylight, and dead fish were removed when noticed. Estimations of LC50 values (concentrations causing 50% mortality in a specified time) were obtained from a computer program for probit (maximum likelihood) and moving-average LC50 computations. Results from both methods agreed within 2%. Abbott's formula (Finney 1971) was used where mortality of control fish occurred.

Daily water samples from each test tank were analyzed for dissolved oxygen using the full-bottle, azide-modified Winkler method (USEPA 1974). The pH, total hardness, and total alkalinity were determined daily on water samples from one tank, and each tank was sampled at least weekly. The pH was measured with a Beckman model 553 pH meter.[1] Total alkalinity was measured by the EDTA titrimetric method, and total hardness was determined potentiometrically (APHA et al. 1971).

Daily water samples from each tank were analyzed for the test metal with a Perkin-Elmer model 403 atomic absorption spectrophotometer. All metal samples were acidified at a rate of 25 ml concentrated nitric acid per liter of sample. Zinc samples were analyzed directly, while cadmium and copper samples were concentrated 20-fold by an evaporation procedure that yielded cadmium recoveries of 101 ± 3% and copper recoveries of 100 ± 8% (mean ± SD). Correction factors were required for cadmium analysis to adjust for matrix effects caused by the concentrated salts. Precision of the cadmium, copper, and zinc analyses were ±0.2, ±0.4, and ±3 μg/liter, respectively. During the copper toxicity test with coho salmon, no metal analyses were performed, but flow rates of copper stock solution and dilution water were checked daily and were always within ±5% of nominal.

Mean weight of coho salmon was about 2.7 kg (steelhead were slightly larger but were not weighed) so that loading factors for 10 and 33 fish per tank were about 5 and 17 g/liter, respectively. However, dynamic loading factors were 1.0 and 3.3 g/liter/day based on replacement flow, and 0.1 and 0.4 g/liter/day based on replacement plus recycle flows. Recommended maximum loading levels for flow-through toxicity tests are 20 g/liter and 2 g/liter/day (Committee on Meth-

[1] Mention of product does not constitute endorsement by EPA.

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During the cadmium toxicity test with steelhead the well pump was inoperable for a 65-h period beginning at 97 h exposure and concluding at 162 h exposure. Water recycle systems maintained the dissolved oxygen and current in the tanks during this period, but no renewal of water or cadmium was possible.

RESULTS AND DISCUSSION

Acute toxicity and water quality data for the adult coho salmon tests are summarized in Table 1. Dissolved oxygen, pH, alkalinity, and hardness values remained generally stable throughout all three coho salmon tests, but water temperatures were appreciably higher (mean 13.7 C) during the copper tests than during the cadmium and zinc tests (mean, 9.4 and 10.0 C, respectively). Zinc and copper toxicity tests were terminated after 96 h exposure, but the cadmium test was continued through 215 h because little mortality had occurred by 96 h. The 96-h LC50 values for copper and zinc were 46 μg/liter and 915 μg/liter, respectively, while the 215-h LC50 for cadmium was 3.7 μg/liter. The two highest cadmium concentrations (15.6 and 8.7 μg/liter) produced nearly identical time-mortality relationships suggesting a plateauing of the concentration-mortality curve such as reported by Ball (1967).

The naturally declining condition of the test fish resulted in 15% mortality of coho salmon in the copper and cadmium toxicity tests. No mortality of control fish occurred in the other four tests.

Toxicity and water quality data from the steelhead tests are also presented in Table 1. The general chemistry of the water supply varied appreciably from day to day during these tests, and as a result, the hardness and alkalinity varied widely within and among toxicity tests. Since metal toxicity varies inversely with these two parameters, this fluctuation undoubtedly caused some irregularity in mortality rates. The 96-h LC50 for zinc was 1,755 μg/liter; the hardness and alkalinity during this toxicity test were the highest of any of the six tests.

The 96-h LC50 for adult steelhead and copper was 57 μg/liter. The mortality curves for steelhead were similar to those for coho salmon in that mortality ceased totally prior to the termination of the 96-h toxicity test. Mortality patterns in acute toxicity tests with copper and adult salmonids typically showed an abrupt decrease in deaths after about 72 h exposure. The cessation of mortality occurred later in the test with steelhead than in the test with coho salmon, and this effect may have been a reflection of the different hardness and alkalinity levels.

During the cadmium toxicity test with adult steelhead, no mortality occurred during the first 200 h of exposure. However, by the termination of the test at 408 h, the LC50 was 5.2 μg/liter. The long period prior to the onset of mortality was perhaps due to the greater hardness and alkalinity, but the 65-h period when the test was essentially a circulating static test may also have contributed to the delay.

The fluctuating hardness and alkalinity which occurred during the adult steelhead tests complicated comparisons between these mortality data and those from the adult coho salmon tests conducted in softer water. However, the adult steelhead 96-h LC50 values can be adjusted to a hardness of 20 mg/liter, using the hardness-mortality relationship obtainable from the data of Brown (1968). The relationship between hardness and copper 48-h LC50 data for rainbow trout (Brown 1968) has been confirmed by Chapman and McCrady (1977) who obtained a nearly identical slope for the hardness-copper toxicity relationship for juvenile chinook salmon (*Oncorhynchus tshawytscha*). Moreover, they found the relationship to hold for both 48-h and 96-h LC50 data and for alkalinity as well as hardness. Thus the hardness-metal toxicity relationships of Brown may reasonably represent a valid means for adjusting acute toxicity data for copper, and perhaps other metals, for salmonids in water with low concentrations of metal-complexing organic compounds (such as WFTS well water). With this hardness adjustment made, direct comparison of the data indicated that adult steelhead were slightly more susceptible to the metals than were adult coho salmon (Table 1).

In order to determine the relative metal sensitivity of adult male and juvenile steelhead, the LC50 data from the adult steelhead toxicity tests were compared to similar LC50 data from toxicity tests with juvenile steelhead which were also conducted at this laboratory (Chapman 1978b, this issue). In all cases these comparisons showed that the adult steelhead had higher LC50 values than the juveniles. Comparison of the acute toxicity data for adult male coho salmon with comparable data for juvenile coho salmon (Chapman, unpublished data) indicated that the adult fish had similar or higher LC50 values than juveniles. These results indicate that the declining condition of adult male fish does not cause them to be more susceptible to the acute lethal effects of metals than are juveniles.

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